

**Report of the Independent Panel on Kikuyugrass (*Pennisetum clandestinum* Hochst.
Ex Chiov) Cultivar 'Whittet'**

**Presented to the Animal and Plant Health Inspection Service
United States Department of Agriculture**

The Panel

On March 16, 1999, USDA-APHIS, assembled a panel of scientific experts to collect and review the scientific literature on kikuyugrass with special emphasis on the cultivar 'Whittet'. The panel consists of scientists with a diverse background including the disciplines of genetics, ecology, weed science, ecosystems management and cultivar development and evaluation.

Scope of the Panel's Work

USDA-APHIS has been asked to determine that a cultivated variety of kikuyugrass (specifically the cultivar 'Whittet') be exempted from the quarantine restrictions which affect kikuyugrass. APHIS conducted an extensive review, but sought additional independent input on this issue before making a decision. APHIS assembled a scientific panel to review the available evidence and make a written report which addresses two main areas:

1. Genetics of kikuyugrass cultivar 'Whittet':
 - a. Is 'Whittet' genetically distinct from other kikuyugrass?
 - b. Is it possible for "genetic drift" or other mechanisms to lead to changes in 'Whittet' which make it more like "unimproved" kikuyugrass with respect to invasiveness?
2. What is the evidence that cultivar 'Whittet' differs from other kikuyugrass in its invasiveness?

I. Introduction

The need to develop productive forage cultivars for the tropical and semitropical regions of the world has resulted in the sustained interest for the majority of this century in grasses such as kikuyugrass. Kikuyugrass was initially introduced into South Africa in 1910 and Australia in 1919 due to its potential as a forage (Mears, 1970). Kikuyugrass is persistent under grazing, has high biomass productivity and forage quality and is resistant to diseases and insects (Wilson, 1968; Mears, 1970; Whitney, 1974; Bogdan, 1977; Wong, 1981; Pearson, et. al., 1985; Oram, 1990).

Pennisetum clandestinum Hochst. Ex Chiov, kikuyugrass, is a C4 creeping perennial with thick and extensive rhizomes and stolons that readily root from nodes (Bogdan, 1977). The short stems and long leaves grow up to 20 cm in length. Flowering shoots, that freely form seed, are shorter than vegetative shoots and are generally hidden beneath a mature sward. Consequently, harvesting seed for commercial use is problematic and establishment has been facilitated using stolons.

Kikuyugrass is a native of highlands and mountains in tropical East Africa: Ethiopia, Eritrea, Kenya, Uganda, Rwanda, Zaire and Tanzania (Edwards, 1937; Mears, 1970). The common name kikuyugrass was derived from the Kikuyu people of Kenya. It grows at altitudes ranging from 1,500 to 3,000 m and rapidly colonizes land at the edges of forests, recently cleared land, roadsides and can become weedy in arable land. Kikuyugrass requires fertile and moist soil (rain fall in its natural range is 1,000 to 1,600 mm per year) in order to thrive. It can tolerate low pH; occasional, mild frost; and drought, but will not survive extended water-logged conditions.

A. Genetic Distinctiveness of cv. Whittet

The development of improved cultivars is dependent on genetic variation within the wild population or a mechanism to artificially create it. The basis of natural variation is sexual reproduction or spontaneous mutations. Plant species that out-cross have greater genetic diversity than species that are self-pollinated or vegetatively propagated. Although the mode of reproduction (and consequently the genetics of this species) of kikuyugrass is not completely understood, several conclusion have been reported (Edwards, 1937; Narayan, 1955; Younger, 1961; Bogdan, 1970; Wilson, 1970; Wilen, 1995). Kikuyugrass reproduces asexually through stolons and rhizomes. Several scientists report apomictic reproduction but this has been refuted by other investigators. The degree to which kikuyugrass self pollinates or out-crosses is still controversial. Several mechanisms are present in this species which increase the chances of out-crossing: male sterile flowers (populations) and; emergence of stigmas 1 to 3 days prior to stamen development.

1. Current Evidence of Genetic Distinction

Genetic diversity was first documented by Edward (1937) who described three naturally occurring ecotypes in Kenya: 'Kabete', 'Molo' and 'Ronga'. Genetic variability in the

native population was sufficient to have allowed the development of several cultivars, including 'Whittet', without selective breeding (Bogdan, 1977). The cultivar 'Whittet' was derived from wild seed collected in 1960 by A. V. Bogdan in Kenya (Oram, 1990). This seed was used to establish 1800 single spaced plants used for observation and an isolated seed increase plot. This accession was designated Mk319 which was later changed to P.713. No variation was observed between the single spaced plants, but these plants in general were noted to be taller, with coarser and wider leaves and more vigorous than accession Mk268 or wild kikuyugrass. 'Whittet' was submitted for registration by the New South Wales Department of Agriculture and registered by the New South Wales Herbage Plant Liaison Committee in 1970. Apparently, after the original collection, there was no systematic selection (other than natural mortality) or artificial cross-pollination in the development of 'Whittet'. Since its release, 'Whittet' has been produced under various conditions and has retained unique characteristics that continue to be in demand by growers (Hosaka, 1958; Wilson and Rumble, 1975; Humphrey, 1980; Kemp, et. al., 1980).

a) Morphological and Phenological Characteristics of the Four Available Cultivars

Whittet - is a taller variety which makes seed harvesting easier. It has broader leaves, higher total crude protein, thicker stems, and larger internodes on the stolons, and it persists better under lower fertility; however it takes longer to form a dense sward. 'Whittet' is uniformly male fertile which enhances seed production and flowers throughout the year.

Crofts - is a taller variety with more upright, narrower leaves than 'Whittet', and more cold tolerant. Although it is susceptible to the Kikuyu yellows, the disease is not prevalent in the cool climates where 'Crofts' is superior.

Breakwell - has fine, narrow leaves and more prostrate runners, forming a dense sward. Although it seeds freely, seed is rarely available now.

Noonan - is very similar to 'Whittet' and 'Breakwell' (its parents), but is recommended for its high field tolerance to Kikuyu yellows, a disease caused by a Phycomycete.

b) Analysis of DNA

To date, there have been no direct DNA analyses of kikuyugrass. However, genetic studies have been conducted including enzyme analysis of kikuyugrass populations in California, and broad- and narrow-sense heritability experiments (Wilén et al., 1995).

Genetic Composition and Potential Drift--Likelihood of Large Population Outcrossings

According to Stebbins (1950) there are many genera that consist of tropical disjuncts from the Old World to the New World. One of the examples Stebbins provides is for the genus *Pennisetum*, where *P. trachyphyllum* of Africa is vicarious to *P. latifolium*, *P.*

bombusaeforme, and *P. tristachyum* of tropical America. According to Stebbins, this vicarious pantropical distribution of species groups was already differentiated by the Eocene.

The genus *Pennisetum* is highly variable in chromosomal numbers with gametophytic numbers ranging from 7 to 34 and sporophyte numbers ranging from 27 to 56 (Goldblatt, 1984). Multiple chromosome numbers have been reported for several taxa, i.e. *P. pedicellatum* and its subspecies with sporophyte numbers of 18, 35+1B, 36, 42, 45, 48, 52, 54 (Brunken, 1979; Chatterji and Sahu, 1980).

The genus *Pennisetum* (Panicoideae; Panicodae; Paniceae) contains about 80 species that are found primarily in tropics and subtropics of both hemispheres (Hitchcock, 1950; Gould, 1975; Wilson and Dallwitz, 1992). Species in this genus possess C_4 anatomy (Wilson and Dallwitz, 1992) common to numerous weedy grass species, e.g. johnsongrass [*Sorghum halepense* (L.) Pers.] and bermudagrass [*Cynodon dactylon* (L.) Pers.]. *Pennisetum* species are commonly adventive and range from helophytic, mesophytic, and xerophytic habitats including a diversity from shaded woods to open savannas.

Hitchcock (1950), Gould (1975), Hickman (1993) and Wunderlin (1997) list the following species of *Pennisetum* in the United States: *P. aepecuroides* (L.) Spreng. sparingly cultivated, escaped in Berks Co., PA, a native of Asia; *P. americanum* (L.) Leeke [= *P. glaucum* (L.) R. Br.], pearl millet, cultivated to some extent in the southern U.S. for forage; *P. cilare* (L.) Link. occasionally cultivated in the southern U.S. and reported to be adventive in wool waste in Yonkers, NY, a native of India; *P. clandestinum* Hochst. ex Chiov., kikuyugrass, [2n=36 (Hrishi, 1952)] an introduced troublesome weed of orchards and gardens in coastal and southern California, a native of Africa; *P. macrostachyum* (Brongn.) Trin. sparingly cultivated ornamental, a native of East Indies; *P. latifolium* Spreng. occasionally cultivated as ornamental, native of South America; *P. nervosum* (Nees) Trin. (2n=36) an introduction from South America; *P. pedicellatum* Trin. [2n=18, 35+1B, 36, 42, 45, 48, 52, 54 (Brunken, 1979; Chatterji and Sahu, 1980)] natives of the Old World; *P. polystachyon* (L.) Schult. [three varieties worldwide with 2n= 34, 36, 45, 54, 56 (Brunken, 1979; Nagabhushana Rao Sindhe, 1980)] a native to tropical America; *P. purpureum* Schumacher, Napier grass or also called elephant grass, introduced and grown as a forage in central to south Florida [2n=28 (Dujardin, 1979)]; *P. setaceum* (Forssk.) Chiov. natives of Africa; *P. setosum* (Swartz) L. Rich. a native to open slopes and savannas of south Florida and tropical America; and *P. villosum* R. Br., feathertop, (2n=18, 27, 36, 45, and 54) cultivated and sparingly escaped in dry soils in California, Michigan, Texas, a native of Africa that is commonly grown as an ornamental and occasionally as an escape from cultivation (naturalized). In addition, Kartesz (1994) lists *P. macrourum* Trin. and *P. petilare* (Hochst.) Chiov. in his checklist of the vascular flora of the United States, Canada, and Greenland. Wagner et al (1990) list *P. clandestinum*, *P. polystachyon*, *P. purpureum* and *P. setaceum* from the Hawaiian Islands. They have this to say about *P. clandestinum*: Kikuyugrass is one of the most serious pest species threatening native vegetation; its smothering, thick, dense growth prevents virtually any new seedling establishment.

There is evidence that genetic diversity occurs within kikuyugrass from its native lands and the introductions into California. Edwards (1937) identified three kikuyugrass cultivars, two cultivars, 'Rongai' and 'Kabete', were determined to be facultative apomicts and that male sterility is recessive to male fertility (Narayan, 1955). From isozyme analysis Wilen et al. (1995) discovered that 12 different genotypes were present in a sample of 354 of the kikuyugrass plants from 3 golf courses in California. Of these, three of nine putative loci varied among genotypes and two of the genotypes occurred at all three locations and represented 73% of the plants sampled. They also determined that, while maintained by colonial reproduction (mowing) and with low genetic variation, kikuyugrass was an aggressive invader through asexual reproduction.

Kikuyugrass cv. 'Whittet' was selected from the genomic diversity within *Pennisetum clandestinum* (Aitkin, 1973). The genetic uniqueness of cv. 'Whittet' is expressed by morphological characteristics that include taller growth habit, slightly broader leaves, thicker stems, longer internodes under spaced plant conditions, and slower growth to form a dense "sward" than the other cultivars of kikuyugrass (Aitkin, 1973; Oram, 1990). In addition to the morphological differences, cv. 'Whittet' is uniformly male fertile (Aitkin, 1973), while the diversity within kikuyugrass includes cultivars that possess hidden and exposed cleistogenes (inflorescences) (Wilson and Dallwitz, 1992). It is possible that cv. 'Whittet' may act as a restorer line and produce fertile progeny when grown in the presence of a male sterile wild strain of kikuyugrass. Younger (1961) demonstrated that male fertile and male sterile crosses of kikuyugrass resulted in fertile offspring that were more competitive (aggressive) than the wild types of kikuyugrass in California. If the same were true with crosses between the progeny of wild strains with cv. 'Whittet' and if most of the morphological traits from cv. 'Whittet' were retained, the resulting progeny could be as or more aggressive than either parental type. Additional research is needed to determine the genetic composition and morphological expression of traits in hybrids between the wild kikuyugrass and cv. 'Whittet'.

Intraspecific and Interspecific Hybridization Within Kikuyugrass and Other Warm Season Grasses

Intraspecific hybridization readily occurs between biotypes, ecotypes, and morphotypes of several grasses. Bermudagrass and johnsongrass are both warm season perennial grasses that are native to the Old World; are able to reproduce sexually (seeds) or asexually (rhizomes or stolons); and are among the World's worst weeds (Holm et al, 1977). Within each of these taxa there is enough genetic variability, natural selection pressures, and intraspecific hybridization that many biotypes or ecotypes have arisen (Rochecouste, 1962a; Rochecouste, 1962b; McWhorter, 1971; Bryson and Wills, 1985; Bryson 1990). These natural selections have resulted in plants that may or may not differ in habit, morphology, and habitat requirements. Likewise, these biotypes or ecotypes may or may not vary in growth and spread potential alone or in competition with agronomic crops (Rochecouste, 1962a; McWhorter 1971; Bryson, 1990). In addition, these biotypes or ecotypes vary in susceptibility or tolerance to herbicides (Rochecouste, 1962b; Hamilton and Tucker, 1964; McWhorter 1971; Bryson and Wills, 1985). Once

established, biotypes or ecotypes of bermudagrass and johnsongrass are able to compete and spread vegetatively or by seeds and fill niches with differing environmental and edaphic conditions. Intraspecific hybridization has been reported in kikuyugrass (Younger, 1961).

Interspecific hybridization between johnsongrass and sorghum [*Sorghum bicolor* (L.) Moench] has resulted in a myriad of phenotypic expressions (de Wit, 1978; Yatskievych 1999). The plant called johnsongrass in the U.S. is probably a stabilized hybrid from *S. bicolor* and *S. halepense*, because true *S. halepense* differs morphologically in its natural habitats within the native range. Currently, there is no evidence that kikuyugrass, including cv. 'Whittet', will or will not hybridize with other species of *Pennisetum*. Simple breeding experiments could determine the potential for interspecific hybridization.

Mutation Potential

There is no evidence from the literature that kikuyugrass, including cv. 'Whittet', has undergone mutations in nature. Natural genetic diversity and selection pressures in the wild and from manipulation by breeders seem to account for all current kikuyugrass selections. If a mutation were to occur in kikuyugrass, including cv. 'Whittet', survival would depend on the mutants ability to survive environmental and edaphic conditions. The rate of spread, like natural or breeder selections, would be dependent on the reproductive potential, type of reproductive system (asexual or sexual), and the vectors for spread.

II. Evidence of Invasive Ability in cv 'Whittet'

Background – Kikuyugrass (*Pennisetum clandestinum* Hochst. Ex Chiov.) is a warm season, strongly rhizomatous and stoloniferous perennial adapted to subtropical and warmer temperate zones with an annual rainfall of 35 inches or more. It is native to the highlands of Central East Africa at altitudes of 6,000 to 10,000 feet. It occurs from Eritrea, through Ethiopia, Kenya, Uganda, Tanzania and Mosambique. Edwards (1937) described two fertile ecotypes which he named 'Kabete' and 'Molo', and a male sterile ecotype which he named 'Rongai'. Narayan (1955) later demonstrated the basis of the fertility in 'Kabete' and the sterility in 'Rongai'.

In 1919, a small seed sample from the Belgian Congo was sown at the Botanic Gardens in Sydney, Australia (Aitken (1973), Sauer) and freely distributed throughout adapted areas in Australia. In 1966, Graeme Wilson screened the Australian collections from Kenya and selected the most fertile plants (Aitkens, 1973). In 1969, the cultivar 'Whittet' was released (Aitkens, 1973). In 1970, 'Whittet' was registered as a commercial cultivar in Australia (Register of Australian Herbage Cultivars, 1990). By 1990, 'Breakwell', 'Crofts', and 'Noonan' also were registered as kikuyugrass cultivars.

A. Current Evidence of Invasive Ability in Comparison With Unimproved Wild Type Kikuyugrass

'Whittet' was selected as an aggressive forage cultivar. The originator (Aitkins, 1973) and the Register of Australian Herbage Cultivars (1990) describe 'Whittet' to be taller, coarser, with broader leaves, thicker stems, longer internodes, and more vigorous than common kikuyugrass. Additionally, 'Whittet' is uniformly fertile. In Australian trials on various soil types (clay loam, sandy loam, shale, sandstone and granite), 'Whittet' was more productive and survived better under conditions of lower or declining soil fertility than common kikuyugrass (Wilson, 1970). This is most likely due to the more highly developed root system of 'Whittet' compared to common kikuyugrass.

B. Evidence of Invasive Ability Under Varying Environmental/Climatic Conditions

Kikuyugrass has been introduced into many tropical and subtropical areas of the world. The seeded varieties appear to be more cold tolerant and may have a more extended range. In Australia, kikuyugrass is widely naturalized down the east coast, across parts of southern Australia and parts of Tasmania (Randall, 1999). In the United States, Kikuyu grass is naturalized in Hawaii (Hosaka, 1958), in California from San Francisco to the Mexico boarder (Wilen et al., 1995; Youngner, 1961), and in lesser areas in Arizona and Texas.

In its naturalized state, kikuyugrass can be found in sterile stands or mixed stands of both sterile and fertile ecotypes (Youngner, 1961). In this state, stolons are frequently found climbing fences, trees and shrubs and rapidly crowding out other species (Hosaka, 1958; Youngner, 1961). In established pastures, it is frequently used to eliminate unwanted weeds such as *Eupatorium*, *Psidium*, *Lantana* (Hosaka, 1958). In highly managed stands with various mixtures of clover or other grasses, care or annual overseeding is required to keep kikuyugrass from taking over as a pure stand (Humphreys, 1980; O'Reilly, 1981; Osaka, 1958; Sauers, ()).

While 'Whittet' has not been tested in competing with the common type, information is available on fertile vs non-fertile ecotype interaction (Youngner, 1961). In stands where both ecotypes were found, seldom were they mixed, except at the periphery where patches of the two met. Most stands consisted of both ecotypes growing in well defined patches. In mixed stands of the two types, the fertile type appears to completely resist invasion by the sterile type. New patches of sterile ecotypes are rare. However, new patches around fertile ecotypes were abundant and extended for some distance around the primary stand.

There is currently no literature on the comparison of climatic effects between 'Whittet' and common kikuyugrass. However, newer selections of fertile cultivars such as 'Crofts' exhibit greater cold tolerance than previous cultivars or common kikuyugrass (Sauers, ()). A climate model (Randall, 1999) predicted that within the US, kikuyugrass would be adapted from California across the Gulf states and Florida up into Georgia and South Carolina. Additionally, kikuyugrass could be adapted to milder areas of Washington and Oregon which are similar in climate to the cooler parts of Tasmania. The greater cold

tolerance of newer cultivars would allow these to out compete common kikuyugrass in the cooler environments.

C. Evidence Under Different Management Regimes

Kikuyugrass is managed as either a turf or forage. Flower and seed formation is stimulated by mowing (Wilson, 1970). Under closely managed turf conditions, such as golf courses, public parks or lawns it is unlikely that kikuyugrass would flower. Spread of kikuyugrass would be mainly by stolon. However, at the peripheral edge or in higher cut fairway rough, there may be a chance for escape and flowering (Wilen et al, 1995).

As stated above, flowering is stimulated by mowing or grazing. This increases the chances of 'Whittet' being spread by animals. Some of the ripe seed taken in by grazing animals will pass out unharmed in the dung (Williams et al., 1975). If these cows were later turned into a pasture of some other forage, the chance of spread is likely.

In pastures which are not heavily grazed, seed could lay dormant until the opportunity to germinate occurred. Seedlings from new seed planted immediately after collection appeared in 69 days (Youngner, 1961). After approximately six months, 94% of the seed had germinated. Seed collected from partially decomposed mats (thatch) germinated in five weeks. It was estimated from the thickness of the mats that the seed may have been three to five years old (Youngner, 1961). Three features favor survival of kikuyugrass under unfavorable conditions: the long dormancy period, uneven germination, and viability for up to five years.

Conclusions

I Genetics of kikuyugrass cultivar 'Whittet'

a. Is 'Whittet' genetically distinct from other kikuyugrass?

1. Wild populations of kikuyugrass had sufficient variation to have allowed for the development of several wild ecotypes and registered cultivars. In addition, 'Whittet' has been used commercially since its release in 1970 and continues to perform under different conditions around the world to the satisfaction of many growers. The evidence strongly supports the conclusion that kikuyugrass has significant genetic diversity and 'Whittet' has demonstrable genetic distinctiveness.
2. Studies could be conducted using molecular techniques to substantiate the genetic distinction of 'Whittet'. These analyses would determine if 'Whittet' is distinct from wild kikuyugrass and other cultivars and how much genetic diversity exists within the cultivar (ie., what is the potential for adaptation to various conditions if it escaped cultivation).

b. Is it possible for “genetic drift” or other mechanisms to lead to changes in ‘Whittet’ which make it more like “unimproved” kikuyugrass with respect to invasiveness?

1. Additional research is needed to determine the genetic composition and morphological expression of traits in hybrids between the wild kikuyugrass and cv. ‘Whittet’. Simple breeding experiments could determine the potential for interspecific hybridization.
2. Studies could be conducted under controlled and field conditions to ascertain the genetic potential of ‘Whittet’ (ie., genotypes within the cultivar) to adapt (if some plants escaped cultivation) to climatic conditions outside its natural range of conditions.

II What is the evidence that the cultivar ‘Whittet’ differs from other kikuyugrass in its invasiveness?

The panel could not find any literature which directly addressed studies utilizing the cultivar ‘Whittet’. However, there are studies which utilize other seeded cultivars of kikuyugrass or seeded wild types of kikuyugrass. These conclusions are based on these reports.

1. Seeded kikuyugrass has been demonstrated to pass through the gut of grazing animals and spread to other pastures in the dung.
2. Reports on the internet involving seeded kikuyugrass have reported that it is a very difficult grass to manage. The reports indicate that “it is an aggressive invader, spreading by way of stolons, rhizomes and seed, with the help of mowers, golfers, football players and anything that is dragged, carried or rolled on the grass. In infested regions, low-maintenance turf facilities such as parks and home lawns eventually convert to kikuyugrass, regardless of the species in the original sward.” Additionally, “seeds can ride on wet shoes, equipment and in soil and may be able to become established in relatively open areas.” Once established on golf courses, it often invades greens and can only be removed by hand weeding. “In ornamental areas, it invades ground covers and flower beds, often completely choking them out.” “In orchards it can compete with trees for nutrients and interfere with irrigation by blocking sprinkler heads and emitters.”
3. Three features favor the survival of kikuyugrass under unfavorable conditions: 1) a long dormancy period, 2) uneven seed germination, 3) viability of seed in thatch for up to five years.
4. Newer seed reproducing cultivars, such as ‘Whittet’, exhibit greater cold tolerance and would allow these cultivars to spread outside the predicted range or to out compete common kikuyugrass in cooler environments.

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